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(54) Title: METHOD FOR EXTENDING THE VASCULAR DWELL-TIME OF PARTICULATE THERAPEUTIC AND PARTICULATE DIAGNOSTIC AGENTS

(57) Abstract

The present invention relates to a composition for increasing the vascular dwell-time of a particulate therapeutic or diagnostic agent in a mammal through the adjunct administration of an effective amount of the particulate therapeutic or diagnostic agent and an effective amount of a vascular dwell-time enhancing agent. Suitable therapeutic and diagnostic agents include those whose efficacy within the body is predicated on their ability to remain within or to be carried within the vascular compartment. Accordingly this method can be readily adapted for treating several diseases and disorders, including hypoxia, hypoxemia, anemia and cancer and for imaging selected regions of a mammal by various imaging techniques, including ultrasound imaging, X-ray imaging and MRI imaging.

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METHOD FOR EXTENDING THE VASCULAR DWELL-TIME  
OF PARTICULATE THERAPEUTIC AND  
PARTICULATE DIAGNOSTIC AGENTS

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1. TECHNICAL FIELD

The present invention relates to a method for increasing the vascular dwell-time of a particulate therapeutic or particulate diagnostic agent in a mammal through the adjunct administration of an effective amount of the particulate therapeutic or particulate diagnostic agent and an effective amount of a vascular dwell-time enhancing agent. This method, therefore, provides a means by which a therapeutic or diagnostic agent is capable of expressing its efficacy by remaining within the vascular compartment or being carried within the vascular compartment to a target. Accordingly, this method is particularly well-suited for treating several conditions in a mammal, including hypoxia, hypoxemia, anemia, infection and cancer as well for imaging selected regions within a mammal by various imaging techniques, including ultrasound imaging, X-ray imaging and MRI.

25       2. BACKGROUND OF THE INVENTION

The efficacy of many therapeutic and diagnostic agents is predicated on the ability of these agents to be retained within the vascular compartment or to be carried within the vascular compartment to their target within a mammal. Unfortunately, many of these agents when they are within the vascular compartment appear particulate in size, as this concept is defined below in Section 4.1. Because these agents activate or prime phagocytes (phagocytic cells) that are present within the vascular compartment and in fixed tissues of the body, they are susceptible to phagocytosis and pinocytosis. The triggering of these two processes in turn results in the accelerated removal of these agents from the vascular

compartment. These agents are then stored within the organs of the reticuloendothelial system, as well as other tissues, until they are metabolized or excreted.

5 In this context, the length of time during which these agents are within the vascular compartment, that is, their vascular dwell-time (intravascular persistence), is relatively short. Because these agents display short dwell-times within the vascular compartment, the ability 10 of these agents to express their efficacy is severely compromised.

Furthermore, when the phagocytic response induced by these agents is particularly pronounced, the phagocytes that are present within the vascular compartment and the fixed tissues of the body often trigger a number of secondary responses which, in turn, cause a number of negative side-effects. For example, activation of phagocytes is known to cause the release of cytokines, such as Tumor Necrosis Factor (TNF) and 15 Interleukin I, as well as ecoisanoids, such as Thromboxane A<sub>2</sub> and Prostaglandin E<sub>2</sub>.

These cytokines and ecoisanoids can cause fever, back pain and pulmonary hypertension. See Inflammation: Basic Principles and Clinical Correlates 20 (J.I. Gallin, I.M. Goldstein and R. Snyderman eds. 1988). Similarly, it has been observed that mammals that have received a particulate fluorocarbon suffer from cachexia. Cachexia is the name given to a generally weakened condition of the body or mind resulting from any 25 debilitating chronic disease. Typical symptoms of cachexia include severe weight loss, depression, loss of appetite, anorexia and anemia. Cachexia is normally associated with neoplastic diseases, chronic infectious diseases or thyroiditis, and is a particular problem when 30 associated with cancerous conditions. In particular, 35 cachexia often compromises a mammal's response to chemotherapy and radiotherapy. This condition, in particular, is believed to be induced by the secretion of

the cytokine TNF. See Tracey, K.J. et al.  
"Cachectin/Tumor Necrosis Factor Induces Cachexia,  
Anemia, Inflammation," J. Exp. Med. 167 :1211-1227  
5 (1988).

Traditionally, the obstacles posed by increased phagocytic activity have been overcome by the sophisticated regulation of a number of parameters, including the regulation of the size of these agents, the 10 dosage employed, the rate of infusion and the frequency with which these agents are administered.

In this context, there is need within the art to extend the vascular dwell-time of these agents and, therefore, counter the effects of phagocytosis, by 15 methods that are not predicated on the intricate regulation of the aforementioned parameters.

### 3. SUMMARY OF INVENTION

The instant invention relates to a novel method 20 by which the vascular dwell-time of a particulate therapeutic or particulate diagnostic agent can be enhanced. The method comprises the adjunct administration of a particulate therapeutic or particulate diagnostic agent and a vascular dwell-time 25 enhancing agent, as the term is defined in Section 4.1., to a mammal.

Accordingly, the invention relates to a method for delivering a particulate therapeutic or particulate diagnostic agent by means of the vascular compartment of 30 a mammal which comprises the adjunct administration of an effective amount of the particulate therapeutic or particulate diagnostic agent and an effective amount of a vascular dwell-time enhancing agent to a mammal.

The present invention further relates to a 35 method for preventing or treating several mammalian conditions, in particular, hypoxia, hypoxemia and anemia. This method comprises the adjunct administration of a therapeutically effective amount of a particulate blood

substitute and an effective amount of a vascular dwell-time enhancing agent to a mammal in need of said prevention or treatment.

5       The present invention also relates to a method for treating a neoplastic condition in a mammal which comprises irradiating a tumor cell in the mammal at least about three hours after the adjunct administration of a therapeutically effective amount of a particulate blood 10 substitute and an effective amount of a vascular dwell-time enhancing agent to the mammal.

15     Furthermore, the present invention relates to a method for imaging a selected tissue or organ of a mammal which comprises imaging the tissue or organ after the mammal has received an adjunct administration of an effective amount of a particulate imaging agent and an effective amount of a vascular dwell-time enhancing agent.

20     In addition, the present invention relates to a method for preventing or treating cachexia in a mammal which comprises the administration of an anti-cachexia agent to a mammal in need of said prevention or treatment.

25     The present invention also relates to a composition comprising a particulate therapeutic or a particulate diagnostic agent and a vascular dwell-time enhancing agent.

Finally, the present invention relates to a kit comprising:

- 30      1)    a vascular dwell-time enhancing agent associated with a carrier system; and  
          2)    either a particulate therapeutic agent or a particulate diagnostic agent.

4. DETAILED DESCRIPTION OF THE INVENTION

4.1 THE METHOD OF THE INVENTION

Based on the adjunct administration of the  
5 vascular dwell-time enhancing agents of the instant  
invention, the instant invention provides greater  
flexibility in counteracting the effects of phagocytosis  
and pinocytosis. In particular, the traditional  
approaches employed for regulating these effects (the  
10 size of the particulate therapeutic or particulate  
diagnostic agent, the dosage employed, the rate of  
infusion as well as the frequency with which the  
particulate therapeutic or particulate diagnostic agent  
is administered) have taken on diminished importance with  
15 respect to preventing the activation of phagocytes.

In fact, based on the manner in which  
phagocytosis and pinocytosis are suppressed by these  
vascular dwell-time enhancing agents, it is believed that  
greater dosages of the particulate therapeutic and  
20 particulate diagnostic agent can be administered over a  
shorter period of time than could be previously achieved.

In addition, it is believed that once the effects of  
these vascular dwell-time enhancing agents wear off, a  
heightened phagocytic response does not ensue. As a  
25 result, it is believed that vascular dwell-time enhancing  
agents may be affecting the metabolic action of cytokines  
and eicosanoids, as well as the onset of the negative  
side effects they trigger.

Finally, when such vascular dwell-time  
30 enhancing agents are adjunctively administered with a  
particulate therapeutic or particulate diagnostic agent,  
the biodistribution as well as the biodegradability of  
the latter may be significantly enhanced. For example,  
when the particulate therapeutic or particulate  
35 diagnostic agent is a fluorocarbon, it has been found  
that significantly lower levels of the particulate  
fluorocarbon accumulate within the organs of the  
reticuloendothelial system (the liver, spleen, lung and

bone marrow) until the particulate fluorocarbon is metabolized or excreted. Instead, based on the adjunct administration of a vascular dwell-time enhancing agent, it has been discovered that greater levels of the particulate fluorocarbon remain within the vascular compartment. Eventually, the particulate fluorocarbon is excreted through the lungs after the particulate fluorocarbon has served its intended function.

While not being bound by theory, it is believed that the beneficial effects of these vascular dwell-time enhancing agents relate to the manner in which carrier systems, to which the therapeutic and diagnostic agents are associated, are metabolized. Often therapeutic and diagnostic agents are formulated in carrier systems, such as lipids, lipid emulsions or liposomal systems before they are administered in order for example to, increase the tolerance of the therapeutic or diagnostic agent within the vascular compartment. In addition, it is believed that the body will often associate the therapeutic and diagnostic agents with a carrier system once the therapeutic and diagnostic agents are administered. In fact, this phenomenon can occur regardless of whether the therapeutic or diagnostic agents are formulated with a carrier system prior to administration.

In this context, it is the carrier system to which the therapeutic or diagnostic agents are associated after administration that renders the therapeutic or diagnostic agents susceptible to phagocytosis and macrophage activity. Accordingly, it is believed that the vascular dwell-time enhancing agents protect the carrier system to which the therapeutic or diagnostic agents are associated after administration from being metabolized or that the vascular dwell-time enhancing agents activate those lipid metabolic processes that, in turn, enhance the stability of the carrier system to which the therapeutic or diagnostic agents are associated.

after administration. Thus, the enhanced stability of the carrier system, in turn, renders the therapeutic and diagnostic agents less susceptible to phagocytosis and 5 macrophage activity.

Alternatively, it is believed that the vascular dwell-time enhancing agents may correct the abnormal metabolism of the carrier systems, which often results from the production of cytokines or ecoisanoids. For 10 example, one possible mechanism of action could be the down-regulation of cytokine and ecoisanoid production.

A particularly likely theory is that the increase in vascular dwell-time is related to the ability of the vascular dwell-time enhancing agents to inhibit 15 the activity of at least one of the following enzymes: NAD glycohydrolase (NADase), ADP-ribose-synthetase or ADP-ribose-polymerase. The latter two enzymes are believed to repair cells attacked by cytokines such as TNF. This hypothesis has been supported experimentally. 20 In particular, it has been found that nicotinamide, a compound that is believed not only to inhibit activity of the three aforementioned enzymes but also to stimulate cell repair in damaged cells, is useful in preventing and treating cachexia. Thus, agents that inhibit NADase, 25 ADP-ribose-synthetase or ADP-ribose polymerase activity can be used not only to increase vascular dwell-time but also to treat or prevent cachexia.

In this vein, suitable particulate therapeutic and particulate diagnostic agents of the instant 30 invention are those therapeutics and diagnostics which, given the form in which they are administered to a mammal, activate the phagocytes present within the vascular compartment or in the fixed tissues of the body and in turn are cleared from the vascular compartment by 35 phagocytosis or pinocytosis.

The preferred mammal of interest, of course, is the human. Nonetheless, other preferred mammals include

domesticated mammals such as equine, bovine, ovine, porcine, canine, feline, feline and murine species.

The terms "particulate therapeutic" and "particulate diagnostic," refer to any therapeutic and diagnostic agent that is known or to be developed and that, after being administered, would be capable of activating the phagocytes of the vascular compartment or the fixed tissues of the body. Accordingly, these two terms refer to those therapeutic and diagnostic agents that, on their own are capable of activating these phagocytes. In addition, these two terms refer to those agents that, because of their association with a carrier system within the body, would be capable of activating these phagocytes. Typically, these agents measure from about .02 to about 10 microns in diameter and more typically from about .05 microns to about 1 micron in diameter. Generally speaking, phagocytes are activated when a particle within the vascular compartment measures from about .001 to about 50 microns in diameter.

Suitable carrier systems to which the therapeutic or diagnostic agents can be associated include lipids, liposomes, microspheres, molecules to which the therapeutic or diagnostic agent is conjugated or cross-linked, artificial cells, microcapsules, microcarrier beads as well as any other carrier system known to one of ordinary skill in the art or to be developed in the future, so long as the result is a particulate therapeutic agent or a particulate diagnostic agent.

With respect to suitable vascular dwell-time enhancing agents, it has been surprisingly discovered that lipid metabolism modifying agents, certain antibiotics, cyclooxygenase inhibitors, NADase inhibitors, ADP-ribose-polymerase inhibitors and ADP-ribose-synthetase inhibitors are capable of extending the vascular dwell-time of a particulate therapeutic or a particulate diagnostic agent so that the latter class of

agents is, in turn, more capable of expressing its efficacy.

The term "lipid metabolism modifying agents" 5 refers to those agents which, when introduced to a mammal, are capable of modulating the synthesis or activity of at least one key lipogenic enzyme. A lipogenic enzyme is an enzyme that is capable of influencing the generation of lipids in fats. Non- 10 limiting examples of such enzymes include lipoprotein lipase, membrane lipase, phospholipase A<sub>2</sub>, and 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase. Accordingly, any agent that is known or to be developed and which falls within this definition can be utilized in 15 the instant invention.

Non-limiting examples of lipid metabolism modifying agents include: clofibrate; gemfibrizol; probucol; thyroxin; insulin; lovastatin; nicotinic acid; and nicotinamide (niacinamide). Preferred lipid 20 metabolism modifying agents are nicotinamide and nicotinic acid, with nicotinamide being more preferred.

Suitable antibiotics are those antibiotics known or to be discovered that are derived from the following sources: *Streptomyces*, *Bacillus subtilis*, 25 *Cephalosporin acremonium*, as well as variants of *Micromonospora purpurea* and *Micromonospora echinospora*.

Non-limiting examples of antibiotics derived from *Streptomyces* include neomycin, streptomycin, kanamycin, paromomycin, novobiocin, vancomycin, 30 lincomycin, oleandomycin, chloramphenicol, clindamycin, cefoxitin, amphotericin B, tobramycin and amikacin. A non-limiting example of an antibiotic derived from *Bacillus subtilis* is bacitracin. Non-limiting examples of antibiotics derived from *Cephalosporin acremonium* 35 include: cephalosporin C and semisynthetic antibiotic derivatives of cephalosporin C. A non-limiting example of an antibiotic derived from *Micromonospora purpurea*, *Micromonospora echinospora* or a variant of these two

strains is gentamicin. Of these antibiotics, the preferred antibiotic is neomycin. It is important to note that the invention does not require that these 5 antibiotics be derived from the aforementioned natural sources. As a result, the invention encompasses these antibiotics regardless of how they are made.

The vascular dwell-time enhancing agent can also be any cyclooxygenase inhibitor, known or to be 10 discovered in the future, with the preferred cyclooxygenase inhibitor being ibuprofen.

Of all these classes of vascular dwell-time enhancing agents, however, the lipid metabolism modifying agents and the antibiotics are preferred, with the lipid 15 metabolism modifying agents being more preferred.

Other non-limiting examples of compounds which are useful as vascular dwell-time enhancing agents include: thymine, thymine analogs such as thymine riboside and thymidine, theophylline, benzamide, as well 20 as benzamide analogs and nicotinamide analogs such as 3-aminobenzamide, benzoic acid, 3-amino-benzoic acid and  $\alpha$ -amino-3-indolepropionic acid, NAD, NADH, and NADPH.

Because the vascular dwell-time enhancing agents, when administered adjunctively, are capable of 25 extending the vascular dwell-time of a particulate therapeutic or a particulate diagnostic agent, the present invention provides an advantageous method for delivering a particulate therapeutic or a particulate diagnostic by means of the vascular compartment of a 30 mammal.

For example, the vascular dwell-time enhancing agents can be instrumental to ensuring that particulate therapeutic agents, such as particulate antibiotics, particulate chemotherapeutics, particulate vasopressors, 35 particulate anti-inflammatory agents, particulate sedatives, particulate anesthetics, particulate tranquilizers, particulate soporifics and particulate vasodilators, and particulate diagnostics, such as

particulate antibodies and particulate radiolabelled compounds, are more capable of exhibiting their efficacy.

In a preferred embodiment, the vascular dwell-time enhancing agents are adjunctively administered with a particulate blood substitute in order to treat conditions such as anemia, hypoxia or hypoxemia. These conditions are often brought about by disorders such as hemorrhagic shock, sepsis, trauma and myocardial infarction.

In addition, the vascular dwell-time enhancing agents of the present invention can be administered with a particulate blood substitute to prevent the onset of these conditions. In this context, the administration of the particulate blood substitute has a prophylactic effect whereby hypoxia, hypoxemia and anemia are prevented. Accordingly, the vascular dwell-time enhancing agent and particulate blood substitute can be administered during any surgical procedure in which blood circulation and perfusion are compromised. Such procedures include angioplasty as well as cardiopulmonary bypass surgery. Thus, in appropriate circumstances, the two agents can be administered via a cell sorting machine or heart-lung machine pump.

Similarly, in another embodiment of the invention, the adjunctive administration of a vascular dwell-time enhancing agent and a particulate blood substitute is incorporated into a regime designed to treat a neoplastic condition. Based on the increased dwell-time of the particulate blood substitute within the vascular compartment, neoplastic tissue has a greater chance of uptaking oxygen. This phenomenon, in turn, renders such tissue vulnerable to the effects of radiation therapy, chemotherapy and hyperthermia therapy.

In fact, the effects of these agents are so great that a mammal can receive multiple treatments of the therapy based on a single dosage of the particulate blood substitute. Accordingly, the need to re-administer

the particulate blood substitute for subsequent treatments is unnecessary. Furthermore, the instant embodiment reduces the toxicological concerns triggered by administering successive dosages of a particulate blood substitute to a mammal.

With respect to radiation treatment, in particular, the effects of the vascular dwell-time enhancing agents are so dramatic that the neoplastic tissue need not receive radiation treatment directly following the adjunct administration of the two agents. Typically, radiation treatment can be administered at least about 3 hours after the adjunct administration of the two agents. While radiation can even be applied about 72 hours after the adjunct administration of the two agents, it is preferred that radiation treatment commence within about the first 3 hours to about 48 hours of the adjunct administration of the two agents.

For each of the embodiments discussed in this Section relating to blood substitutes, any particulate blood substitute known or to be developed can be utilized. Non-limiting examples of particulate blood substitutes include hemoglobin based compositions, such as hemoglobin that has been cross-linked or otherwise polymerized as well as hemoglobin that has been formulated prior to administration with a carrier system, as the latter term has been previously defined in this Section.

In addition, a fluorocarbon, which includes any compound containing a fluorocarbon hydrocarbon moiety, can be used as a blood substitute. Non-limiting examples of fluorocarbons include:

- 1) a bis(F-alkyl)ethanes, such as  $C_4F_9CH=CHC_4F_9$ , (F-44E),  $i-C_3F_7CH=CHC_6F_{13}$  (F-i36E) and  $C_6F_{13}CH=CHC_6F_{13}$  (F-66E),  $C_3F_7CBr=CBrC_3F_7$ ,  $C_3F_7Cl=CIC_3F_7$ ,  $C_2F_5CBR=CBrC_2F_5$ ,  $C_2F_5Cl=CIC_2F_5$ ;
- 2) cyclic fluorocarbons, such as  $C_{10}F_{18}$  (FDC), F-adamantane (FA), F-methyladamantane (FMA),

- F-1,3-dimethyladamantane (FDMA), F-di- or  
F-trimethylbicyclo[3,3,1]nonane (nonane);  
3) perfluorinated amines, such as  
5 F-tripropylamine (FTPA) F-tributylamine (FTBA),  
F-4-methylcytlydroquinolizine (FMOQ),  
F-n-methyl-decahydroisoquinoline (FMIQ),  
F-n-methyldecahydroquinoline (FHQ),  
F-n-cyclohexylpyrrolidine (FCHP) and  
10 F-2-butyltetrahydrofuran (FC-75 or RM101);  
4) halogenated perfluorocarbons, such as:  
i) monobrominated perfluorocarbons, such as 1-bromoheptadecaflourooctane (PFOB),  
1-bromopentadecaflouroheptane, 1-bromotridecafluorohexane  
15 (PFHB);  
ii) dibrominated perfluorocarbons, such as C<sub>6</sub>F<sub>12</sub>Br<sub>2</sub>, C<sub>7</sub>F<sub>14</sub>Br<sub>2</sub> and C<sub>8</sub>F<sub>16</sub>Br<sub>2</sub>;  
iii) monoiodinated perfluorocarbons;  
iv) di-iodinated perfluorocarbons;  
20 5) perfluoroalkylated ethers or polyethers,  
such as (CF<sub>3</sub>)<sub>2</sub>CFO(CF<sub>2</sub>CF<sub>2</sub>)<sub>2</sub>OCF(CF<sub>3</sub>)<sub>2</sub>, (CF<sub>3</sub>)<sub>2</sub>CFO(CF<sub>2</sub>CF<sub>2</sub>)<sub>3</sub>OCF(CF<sub>3</sub>)<sub>2</sub>,  
(CF<sub>3</sub>)<sub>2</sub>CFO(CF<sub>2</sub>CF<sub>2</sub>)<sub>2</sub>F, (CF<sub>3</sub>)<sub>2</sub>CFO(CF<sub>2</sub>CF<sub>2</sub>)<sub>3</sub>F, (C<sub>6</sub>F<sub>13</sub>)<sub>2</sub>O and  
F[CF(CF)CFO]CHFCF;  
25 6) perfluorocyclic ethers;  
7) perfluoroalkylated hydrides, such as  
perfluoroctylhydride, perfluorononylhydride; and  
perfluorodecylhydride; and  
8) fluorocarbon-hydrocarbons, such as C<sub>8</sub>F<sub>17</sub>C<sub>2</sub>H<sub>5</sub>,  
C<sub>6</sub>F<sub>13</sub>CH=CH<sub>2</sub>H<sub>13</sub> and C<sub>8</sub>F<sub>17</sub>R, wherein R is any hydrocarbon with  
30 an organic functional group.

In another embodiment of the present invention, a vascular dwell-time enhancing agent can be administered adjunctively with a particulate contrast agent to image a particular tissue or organ of interest. Non-limiting examples of such imaging techniques include ultrasound imaging, X-ray imaging (such as computed axial tomography imaging (CAT)), and magnetic resonance imaging (MRI).

By virtue of these imaging techniques, valuable diagnostic information can be gathered regarding various tissues, organs and systems within a mammal.

5 Accordingly, these techniques can be used to gather information about the heart, blood as well as the lymphatic system and the vascular compartment. Furthermore, because a sufficient amount of the particulate diagnostic agent still accumulates within the 10 reticuloendothelial system, the liver, lung, spleen and bone marrow of a mammal can also be imaged. With respect to the liver and spleen, these techniques are particularly well suited for detecting tumors and lesions.

15 Suitable particulate ultrasound contrast agents include proteinaceous matrices containing oxygen, air or another gas, such as oxygen embedded albumin microspheres, as well as fluorocarbons, such as those previously recited in this Section.

20 Suitable particulate X-ray contrast agents are compounds that have been labeled with a heavy halogen, such as iodine and bromine, including the monobrominated, dibrominated, monoiodinated and di-iodinated fluorocarbons previously identified in this Section.

25 Finally, suitable particulate MRI contrast agents include: paramagnetic compounds, such as ferric oxyhydroxide, gadolinium oxide, and fluorocarbons, such as those identified previously; ferromagnetic compounds, such as magnetite, ferrite and gamma ferric oxide; and 30 superparamagnetic compounds, such as superparamagnetic iron oxide.

Because the vascular dwell-time of the particulate therapeutic or particulate diagnostic agent is enhanced, the biodistribution and ultimate fate of the 35 agents within the mammal can be dramatically effected as well. For example, with respect to fluorocarbons, greater levels of the fluorocarbons are present in the vascular compartment. Ultimately, the fluorocarbons are

expired across the pulmonary membrane as a gas through the lungs.

Another aspect of the present invention is that  
5 many of the vascular dwell-time enhancing agents, as well as agents that are not vascular dwell-time enhancing agents, can be administered to a mammal in order to prevent or treat cachexia. These agents are termed "anti-cachexia agents." Accordingly, the anti-cachexia  
10 agents can treat or prevent effects conventionally associated with cachexia, such as weight loss, depression, loss of appetite, lipid metabolic disorders, piloerection and decreased responses to chemotherapy and radiotherapy.

15 Suitable anti-cachexia agents include those agents that are known or to be discovered and that are capable of inhibiting NADase, ADP-ribose-synthetase or ADP-ribose-polymerase activity in a mammal. Typically, the anti-cachexia agents also stimulate DNA repair in  
20 damaged cells in a mammal. In this light, a preferred anti-cachexia agent is one that can effect all four of these previously mentioned metabolic mechanisms.

Non-limiting examples of anti-cachexia agents include: thymine, thymine analogs such as thymine  
25 riboside and thymidine, theophylline, nicotinamide, benzamide, as well as, nicotinamide analogs and benzamide analogs such as nicotinic acid, 3-aminobenzamide, benzoic acid, 3-amino-benzoic acid, and  $\alpha$ -amino-3-indolepropionic acid, NAD, NADH and NADPH.

30

#### 4.2. MODE OF ADMINISTRATION

The instant invention, in part, comprises the adjunct administration of a vascular dwell-time enhancing agent and a particulate therapeutic or a particulate  
35 diagnostic agent. In this context, the term "adjunct administration" means that these two agents can be administered to a mammal either as a mixture or, sequentially, so long as the first agent to be

administered is still capable of serving its intended function within the mammal when the second agent is administered.

With respect to the particulate therapeutic or particulate diagnostic agent, this agent can be formulated and administered in accordance with those methods traditionally employed within the art as well as those known to one of ordinary skill in the art. Given the benefits of the vascular dwell-time enhancing agents, however, the conventional dosages and perfusion rates required to counter the effects of phagocytosis may no longer be necessary; for, it is believed that a greater dosage can now be administered over a shorter time. The degree to which adjustments in the dosages and doses to be administration may be required can be readily determined by one of ordinary skill in the art. Accordingly, the term "effective amount of a therapeutic or diagnostic agent" refers to a therapeutically effective amount of a particulate therapeutic agent and a diagnostically effective amount of a particulate diagnostic agent respectively.

Ideally, the vascular dwell-time enhancing agent and the particulate therapeutic or particulate diagnostic agent are packaged in the form of kit which contains an effective amount of a vascular dwell-time enhancing agent and an effective amount of the particulate therapeutic or particulate diagnostic agent. These agents can be packaged so that they can be administered in accordance with any of the regimes and methods mentioned in the following discussion.

With respect to the vascular dwell-time enhancing agents in general, these agents can be formulated together with the particulate therapeutic or particulate diagnostic agent. In addition, these agents can be formulated separately and subsequently mixed with the particulate therapeutic or particulate diagnostic

agent. In either instance, however, both agents can be administered as a mixture.

When the vascular dwell-time enhancing agents  
5 are formulated with the particulate therapeutic or  
particulate diagnostic agent, it is preferred that an  
initial bolus injection of the vascular dwell-time  
enhancing agent be administered to a mammal as a primer.  
This administration is then followed, about 5 minutes to  
10 about 4 hours later, by a second administration,  
comprising a mixture of the vascular dwell-time enhancing  
agent and a particulate therapeutic or particulate  
diagnostic agent.

Generally, when this second administration  
15 contains a particulate therapeutic agent, the mixture is  
preferably administered intravenously over a period of  
time of at least about 15 minutes. Based on the  
particulate therapeutic being administered, the mixture  
may require a longer period of administration. The time  
20 required for administering a given particulate  
therapeutic will be readily appreciated by one of  
ordinary skill in the art.

When the second administration contains a  
particulate diagnostic agent, the mixture is preferably  
25 administered intravenously over a period of time of at  
least about 3 minutes to about 10 minutes. When  
administering either a particulate therapeutic or  
particulate diagnostic agent, additional doses of a  
vascular dwell-time enhancing agent may be administered.  
30 Preferably, these subsequent doses of the vascular dwell-  
time enhancing agent are administered by an intravenous  
infusion, an injection or oral ingestion.

When administered separately, the vascular  
dwell-time enhancing agents and anti-cachexia agents can  
35 be administered enterally, that is, orally, topically, by  
inhalation spray or rectally in dosage unit formulations  
containing conventional non-toxic pharmaceutically  
acceptable carrier systems.

In addition, the agents can be administered parenterally, that is by subcutaneous, intravenous, intramuscular, injection or infusion techniques.

5 As for suitable formulations, the vascular dwell-time enhancing agents and anti-cachexia agents can be formulated in any number of ways, depending on the mode of administration contemplated. For example, when administered orally, the agents can be incorporated into 10 a tablet, troche, lozenge, aqueous or oily suspension, dispersible powder or granule, emulsion, hard or soft capsule, syrup or elixir. Furthermore, these compositions can also contain additional ingredients, such as sweetening agents, flavoring agents, coloring 15 agents or preservatives, in order to provide a more elegant and palatable composition. These compositions can be prepared according to any method known in the art.

When formulated as a tablet, the vascular dwell-time enhancing agents and anti-cachexia agents can 20 be admixed with a non-toxic pharmaceutically acceptable excipient. Suitable excipients are, for example: inert diluents, such as calcium carbonate, sodium carbonate, lactose, calcium phosphate or sodium phosphate; 25 granulating and disintegrating agents, for example, maize starch, or alginic acid; binding agents, for example, starch, gelatin or acacia; and lubricating agents, for example, magnesium stearate, stearic acid or talc.

The tablet can also be uncoated or coated by known techniques in order to delay the disintegration of 30 the tablet and the absorption of the agents by the gastrointestinal tract. As a result, a sustained action over a longer period can be achieved. Suitable time delay agents are glyceryl monostearate and glyceryl distearate.

35 Formulations for oral use can also be presented as gelatin capsules. When in the form of a hard capsule, the vascular dwell-time enhancing agents and anti-cachexia agents are mixed with an inert solid

diluent, for example, calcium carbonate, calcium phosphate or kaolin. As a soft gelatin capsule, the vascular dwell-time enhancing agents are mixed with water 5 or an oil medium, for example, peanut oil, liquid paraffin, or olive oil.

Aqueous suspensions usually contain the vascular dwell-time enhancing agents and anti-cachexia agents in an admixture with appropriate excipients. Such 10 excipients include: suspending agents, for example, sodium carboxymethylcellulose, methylcellulose, hydroxypropylmethylcellulose, sodium alinate, polyvinylpyrrolidone, gum tragacanth and gum acacia; dispersing or wetting agents which can be a naturally 15 occurring phosphatide, for example, lecithin; condensation products of an alkylene oxide with a fatty oxide, for example, polyoxyethylene stearate; condensation products of ethylene oxide with a long chain aliphatic alcohol, for example, 20 heptadecaethyleneoxycetanol; condensation products of ethylene oxide with a partial ester derived from fatty acids and a hexitol such as polyoxyethylene sorbitol monooleate; or condensation products of ethylene oxide with a partial ester derived from fatty acids and hexitol 25 anhydrides, for example, polyoxyethylene sorbitan monooleate.

The aqueous suspensions can also contain additional ingredients, such as: one or more preservatives, for example, ethyl, n-propyl, or 30 p-hydroxybenzoate; coloring agents; flavoring agents; or sweetening agents, such as sucrose or saccharin.

Oily suspensions can be formulated by suspending the vascular dwell-time enhancing agents in a vegetable oil, for example, arachis oil, olive oil, 35 sesame oil or coconut oil; or in a mineral oil, such as liquid paraffin. These suspensions can contain a thickening agent, for example, beeswax, hard paraffin or cetyl alcohol. Sweetening agents, such as those set

forth above, and flavoring agents can be added to provide a palatable oral preparation. Furthermore, these compositions can be preserved by the addition of an antioxidant, such as ascorbic acid.

5 Powders and granules can also be dispersed in an aqueous suspension by the addition of water, a dispersing or wetting agent, and, optionally, one or more preservatives. Suitable dispersing agents, wetting  
10 agents and suspending agents are exemplified by those already mentioned in this Section. Additional excipients, for example, sweetening, flavoring and coloring agents, can also be present.

The vascular dwell-time enhancing agents and anti-cachexia agents can also be prepared in oil-in-water emulsions. The oily phase can be a vegetable oil, for example, olive oil or arachis oils, or a mineral oil, for example, liquid paraffin or mixtures of these. Suitable emulsifying agents can be naturally-occurring gums, for  
15 example: gum acacia or gum tragacanth; naturally-occurring phosphatides, for example, soybean lecithin; esters including partial esters derived from fatty acids and hexitol anhydrides, for example, sorbitan mono-oleate; and condensation products of said partial  
20 esters with ethylene oxide, for example, polyoxyethylene sorbitan mono-oleate. The emulsions can also contain sweetening and flavoring agents.

Syrups and elixirs can be formulated with sweetening agents, for example glycerol, sorbitol or sucrose. Such formulations can contain a demulcent, as  
30 well as a preservative, flavoring agent or coloring agent.

The vascular dwell-time enhancing agents and anti-cachexia agents also can be in the form of sterile injectable aqueous or oleaginous suspensions. These  
35 suspensions can be formulated by those general techniques known in the art and using suitable dispersing or wetting agents and suspending agents which have been

mentioned in this Section. The sterile injectable preparation can be a sterile injectable solution or suspension in a non-toxic parenterally acceptable diluent 5 or solvent. Among the acceptable diluents and solvents that can be employed are water, 1,3-butanediol, Ringers solution and isotonic sodium chloride solution.

In addition, sterile fixed oils are conventionally employed as a solvent or suspending 10 medium. For this purpose any bland fixed oil can be employed including synthetic mono- or diglycerides. Fatty acids such as oleic acid also find use in the preparation of injectables.

The vascular dwell-time enhancing agents and 15 anti-cachexia agents can also be in the form of suppositories for rectal administration. These compositions can be prepared by mixing the agents with a suitable non-irritating excipient which is solid at ordinary temperatures and liquid at the rectal 20 temperature. As a result, the excipient will melt and release the agents in the rectum. Suitable excipients include cocoa butter and polyethylene glycols.

When these agents are administered on their own, they are preferably formulated in a carrier system, 25 non-limiting examples of which have previously been recited in Section 4.1. By administering the vascular dwell-time enhancing agents or anti-cachexia agents in such a manner, one is capable of delivering the agents to specific targets within a mammal, such as the phagocytes 30 present within the vascular compartment or fixed tissue of the mammal. As a result, very high doses of the agents are delivered to the target while very low doses are delivered to the rest of the mammal.

When these agents are not formulated in a 35 carrier system and are administered separately, they are preferably administered in the following manner: when these agents are administered enterally, they are preferably administered orally in 3-4 dosages per day.

When these agents are administered parenterally, they are preferably administered in a manner similar to that discussed previously in this Section. First, an initial bolus injection of these agents is administered. Three to four hours later, the agents are intravenously administered over a 24 hour period. Subsequent 24 hour dosages are repeated until the efficacy of these agents is no longer required.

With respect to the dosage levels to be administered, the dosage level should be generally from about 5 mg/kg per day to about 1,000 mg/kg per day. Ultimately the object of administering these vascular dwell-time enhancing agents and anti-cachexia agents is to maximize either the concentration of the particulate therapeutic agent or particulate diagnostic agent in the blood, or to maximize the appetite and activity of the mammalian subject over a given period of time respectively. At the same time, however, one should strive to apply as low a dose of the vascular dwell-time enhancing agents and anti-cachexia agents as possible and to maintain a constant concentration of these agents within the blood as long as their efficacy is required. Prolonged use of these agents at the lower dosage range recited above should not be problematic given the relatively low toxicity of these agents.

For example, when nicotinamide is administered orally, the dosage should be about 500 mg/kg/day. When nicotinamide is administered by the preferred parenteral mode, an initial bolus injection of about 500 mg/kg should be followed by an intravenous administration of about 500 mg/kg/day.

When neomycin is administered orally, the dosage should be about 30 mg/kg/day to about 50 mg/kg/day. When neomycin is administered by the preferred parenteral mode, an initial bolus injection of about 5 mg/kg should be followed by an intravenous

administration of about 5 mg/kg/day to about 10 mg/kg/day.

When ibuprofen is administered orally, the dosage should be about 10 mg/kg/day to about 40 mg/kg/day. When ibuprofen is administered by the preferred parenteral mode, an initial bolus injection of about 10 mg/kg should be followed by an intravenous administration of about 10 mg/kg/day to 40 mg/kg/day.

When isoniazid is administered orally, the dosage should be about 10 mg/kg/day to about 40 mg/kg/day. When isoniazid is administered by the preferred parenteral mode, an initial bolus injection of about 10 mg/kg should be followed by an intravenous administration of about 10 mg/kg to about 40 mg/kg/day.

When nicotinic acid is administered orally, the dosage should be about 500 mg/kg/day. When nicotinic acid is administered by the preferred parenteral mode, an initial bolus injection of about 500 mg/kg should be followed by an intravenous administration of about 500 mg/kg/day.

## 5. EXAMPLES

### 25 5.1 BIOLOGICAL DATA

#### EXAMPLE I

#### The Effects of Vascular Dwell- Time Enhancing Agents on the Biodistribution of 30 Perfluorooctylbromide (PFOB)

Four experiments were conducted in order to assess the ability of a vascular dwell-time enhancing agent to extend the dwell-time of a particulate therapeutic or diagnostic agent within the vascular compartment. Two experiments focused on the effects of nicotinamide on the concentration of perfluorooctylbromide (PFOB) in various organs of healthy mice as well as mice with either mammary or colon tumors.

The first of these experiments also focused on the ability of neomycin to increase the concentration of PFOB in the blood of mice with mammary tumors. A third experiment compared the effects of nicotinamide with three other vascular dwell-time enhancing agents: isoniazid, nicotinic acid and neomycin. A fourth experiment focused on the effects of ibuprofen on the concentration of PFOB in the blood of healthy mice as well as mice with mammary tumors or Lewis lung tumors.

#### Materials and Methods

##### Mammals:

Female Balb/C normal healthy mice as well as female mice bearing mammary, colon or Lewis lung tumors were employed in these studies. Each mouse weighed approximately 20 gm.

##### Drugs:

Perfluoroctylbromide (PFOB) (with a mean particle size of 250 nanometers) was procured from Alliance Pharmaceutical Corp. and administered intravenously as an emulsion (90 gm/100 ml) in the tail vein.

Nicotinamide (Sigma) isoniazid (Sigma), nicotinic acid (Sigma) neomycin (Sigma) solutions were freshly prepared before each experiment. These compounds were dissolved in water and administered intravenously. Ibuprofen (Upjohn Co.) was obtained as sterile stable solution suitable for intravenous administration.

Normal Saline (0.9% w/v) was administered intravenously in the tail vein.

##### Extraction Techniques:

Blood was thawed and a 50  $\mu$ l sample was removed and mixed with 2 ml of water to lyse the cells. To that sample, 5 ml of ethanol was added in order to break up

the emulsion. Twenty minutes later, 20 ml of isoctane was added to the sample in order to extract the fluorocarbon. The treated sample was then mixed 5 thoroughly and centrifuged at 15000 rpm for 12 min. A sample of the resulting supernatant was then removed for analysis. Tissue samples were subject to a modified version of this protocol, in which, prior to the addition of ethanol, tissue samples were thawed, minced and then 10 homogenized in 2 ml of water.

Gas Chromatography Analysis:

The gas chromatograph was standardized daily by running standard curves with known concentrations of 15 PFOB. Samples were loaded into the machine and subjected to analysis with an electron capture detector. The PFOB concentration was then calculated from the following equation: [PFOB] = (the area under the chromatograph) x (the slope of the standard curve) x (the intercept) x 20 (milliliters solvent per gram of tissue or organ).

**EXPERIMENT 1**

In one experiment, mice were administered intravenously a single dose of PFOB (9 gm/kg). In 25 addition, mice received one of the following: one of three doses of nicotinamide (100 mg/kg, 500 mg/kg, 1000 mg/kg with a rate of perfusion of about 10 ml/kg/min to 20 ml/kg/min); a dose of neomycin (5 mg/kg with a rate of perfusion of about 10 ml/kg/min to 20 ml/kg/min); or a 30 single dose of saline control. The saline control (equivalent in volume to a dose of 1 gm/kg of nicotinamide) was administered by bolus injection or at a rate of perfusion of 10 ml/kg/min. Twenty-four hours post injection, the mice were sacrificed by cervical dislocation. Two milliliters of whole blood were stored 35 in a heparinized tube and frozen (-70°C) until analysis. At least 20 µl, ideally 50 µl, of bone marrow were frozen (-70°C) until analysis. The liver, spleen and lung were

removed in toto and frozen (-70°C). Specimens of adipose tissue were also removed. The results of these experiments appear in Tables I and VI.

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#### EXPERIMENT 2

In a second experiment, mice were administered a single intravenous dose of PFOB (10 g/kg with a rate of perfusion of about 10 ml/kg/min. to 20 ml/kg/min.) and either one, two, or three bolus doses (1 gm/kg each) of nicotinamide intraperitoneally or a single (0.01 ml/kg) dose of normal saline control (as in Experiment 1). Forty-eight hours post injection, the mice were sacrificed by cervical dislocation. The spleen, liver, lung and blood were isolated as in Experiment 1. The results of this experiment appears in Tables VII to IX.

#### EXPERIMENT 3

In a third experiment, mice were administered three bolus doses of a vascular dwell-time enhancing agent intraperitoneally. Nicotinamide was administered at a dose of 500 mg/kg, isoniazid was administered at a dose of 10 mg/kg and neomycin was administered at a dose of 5 mg/kg. Forty-eight hours post injection of PFOB, the mice were sacrificed by cervical dislocation. Blood samples and tissue samples were obtained as in Experiment 1. The results of this experiment appear in Tables X to XIV.

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#### EXPERIMENT 4

In a fourth experiment, mice were administered three bolus doses of ibuprofen (10 mg/kg) intraperitoneally and a single intravenous bolus dose of PFOB (10 g/kg). Forty-eight hours post injection of PFOB, the mice were sacrificed by cervical dislocation. Blood samples were obtained as in Experiment 1. The results of this experiment are summarized in Table XV.

TABLE I. THE CONCENTRATION OF PFOB (MG/G) IN THE BLOOD 24 HOURS AFTER A SINGLE DOSE OF NICOTINAMIDE OR NEOMYCIN.

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
5	SALINE CONTROL	38.53	19.71	3.94	25	
<b>NICOTINAMIDE:</b>						
10	100 MG/KG	34.70	13.77	2.75	25	<.01
	500 MG/KG	37.77	13.82	2.76	25	>0.1
	1000 MG/KG	46.07	19.82	3.96	25	<.01
MICE BEARING MAMMARY TUMORS						
15	SALINE CONTROL	24.36	9.78	2.31	18	
	NICOTINAMIDE (500 MG/KG)	29.45	7.24	1.94	14	<0.05
20	NEOMYCIN (5 MG/KG)	30.45	9.47	2.30	17	+25

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

25 B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY UNDER INVESTIGATION WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE).

TABLE II. THE CONCENTRATION OF PFOB IN THE LIVER 24 HOURS AFTER A SINGLE DOSE OF NICOTINAMIDE.

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	20.38	3.20	0.64	25		
NICOTINAMIDE:						
100 MG/KG	19.06	2.97	0.59	25	>0.1	-7
500 MG/KG	18.28	2.26	0.45	25	<0.005	-10
1000 MG/KG	16.57	2.26	0.45	25	<0.0005	-19

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A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./N

C N REFERS TO THE NUMBER OF MICE TESTED

30 D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY UNDER INVESTIGATION WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE).

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TABLE III. THE CONCENTRATION OF PFOB (MG/G) IN BONE MARROW 24 HOURS AFTER A SINGLE DOSE OF NICOTINAMIDE

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	8.73	5.66	1.13	25		
NICOTINAMIDE:						
100 MG/KG	8.19	5.64	1.13	25	>0.1	-7
500 MG/KG	6.61	4.07	0.81	25	>0.05 < .1	-24
1000 MG/KG	7.30	3.44	0.69	25	>0.1	-16

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A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

30 D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

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TABLE IV. THE CONCENTRATION OF PFOB (MG/G) IN THE SPLEEN 24 HOURS AFTER A SINGLE DOSE OF NICOTINAMIDE.

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NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	%CHANGE
SALINE CONTROL	156.9	14.2	2.8	25		
NICOTINAMIDE:						
100 MG/KG	160.1	13.7	2.8	24	>0.1	+2
500 MG/KG	174.1	15.6	3.1	25	<.0005	+11
1000 MG/KG	177.0	19.7	3.9	25	<0.005	+13

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS.

1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY UNDER INVESTIGATION WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE).

TABLE V. THE CONCENTRATION OF PFOB (MG/G) IN THE LUNG 24 HOURS AFTER A SINGLE DOSE OF NICOTINAMIDE.

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	%CHANGE
SALINE CONTROL	19.5	5.79	1.16	25		
NICOTINAMIDE:						
100 MG/KG	19.97	4.17	0.83	25	>0.1	+2
500 MG/KG	19.52	4.16	0.83	25	>0.1	0
1000 MG/KG	17.84	4.22	0.84	25	>0.1	-8.5

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A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

30 D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

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TABLE VI. THE CONCENTRATION OF PFOB (MG/G) IN ADIPOSE TISSUE 24 HOURS AFTER THE ADMINISTRATION OF PFOB.

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NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	%CHANGE
SALINE CONTROL	4.22	1.66	0.33	25		
NICOTINAMIDE:						
100 MG/KG	5.45	1.91	0.38	25	>0.1	+29
500 MG/KG	4.0	1.54	0.31	25	>0.1	-5
1000 MG/KG	4.56	1.76	0.35	25	>0.1	+8

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A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

30 D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

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TABLE VII. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE CONCENTRATION OF PFOB (MG/G) IN THE BLOOD 48 HOURS AFTER THE ADMINISTRATION OF PFOB

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	14.62	7.34	2.32	10		
NICOTINAMIDE 1X <sup>E</sup>	22.43	4.89	1.65	10	<0.01	+53
NICOTINAMIDE 2X <sup>F</sup>	20.47	3.40	1.08	10	<0.025	+40
NICOTINAMIDE 3X <sup>G</sup>	24.43	4.97	1.57	10	<0.005	+67
MICE BEARING MAMMARY TUMORS						
SALINE CONTROL	0.56	0.51	0.16	10		
NICOTINAMIDE 1X	2.99	3.51	0.78	20	<0.005	+434
NICOTINAMIDE 2X	14.77	6.91	1.55	20	<0.0005	+2538
NICOTINAMIDE 3X	18.06	11.31	2.53	20	<0.0005	+3125

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A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./√N

C N REFERS TO THE NUMBER OF MICE TESTED

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D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE).

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E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

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G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE VII. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE  
 (CONTINUED) CONCENTRATION OF PFOB (MG/G) IN THE BLOOD 48 HOURS AFTER THE  
 ADMINISTRATION OF PFOB

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
5	SALINE CONTROL	14.62	7.34	2.32	10	
10	NICOTINAMIDE 1X <sup>E</sup>	22.43	4.89	1.65	10	<0.01 +53
15	NICOTINAMIDE 2X <sup>F</sup>	20.47	3.40	1.08	10	<0.025 +40
20	NICOTINAMIDE 3X <sup>G</sup>	24.43	4.97	1.57	10	<0.005 +67
MICE BEARING COLON TUMORS						
15	SALINE CONTROL	7.42	6.61	1.55	10	
20	NICOTINAMIDE 1X	5.68	4.4	1.39	10	>0.1 -23
25	NICOTINAMIDE 2X	27.59	12.12	3.83	10	<0.0005 +272
30	NICOTINAMIDE 3X	37.33	17.1	5.41	10	<0.0005 +403

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

25 C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETTLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT

30 T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE).

E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

35 G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE VIII. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE CONCENTRATION OF PFOB (MG/G) IN THE LIVER 48 HOURS AFTER THE ADMINISTRATION OF PFOB

NORMAL HEALTHY MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	26.06	2.89	0.91	10		
NICOTINAMIDE 1X <sup>E</sup>	23.39	2.71	0.86	10	<0.025	-11
NICOTINAMIDE 2X <sup>F</sup>	24.80	2.63	0.83	10	>0.1	-5
NICOTINAMIDE 3X <sup>G</sup>	23.33	2.23	0.71	10	<0.025	-11
MICE BEARING MAMMARY TUMORS						
SALINE CONTROL	39.84	4.12	1.3	20		
NICOTINAMIDE 1X	41.56	6.84	1.53	20	>0.1	+4
NICOTINAMIDE 2X	37.03	3.75	0.84	20	<0.05	-7
NICOTINAMIDE 3X	35.25	4.48	1.00	20	<0.01	-12

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE VIII. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE  
 (CONTINUED) CONCENTRATION OF PFOB (MG/G) IN THE LIVER 48 HOURS AFTER THE  
 ADMINISTRATION OF PFOB

NORMAL HEALTHY MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	26.06	2.89	0.91	10		
NICOTINAMIDE 1X <sup>E</sup>	23.39	2.71	0.86	10	<0.025	-11
NICOTINAMIDE 2X <sup>F</sup>	24.80	2.63	0.83	10	>0.1	-5
NICOTINAMIDE 3X <sup>G</sup>	23.33	2.23	0.71	10	<0.025	-11
MICE BEARING COLON TUMORS						
SALINE CONTROL	36.07	6.89	2.1	10		
NICOTINAMIDE 1X	37.27	4.42	1.40	10	>0.1	+3
NICOTINAMIDE 2X	32.60	3.90	1.23	10	>0.05<0.1	-10
NICOTINAMIDE 3X	28.25	3.39	1.07	10	<0.005	-30

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE IX. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE CONCENTRATION OF PFOB (MG/G) IN THE SPLEEN 48 HOURS AFTER THE ADMINISTRATION OF PFOB

NORMAL HEALTHY MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	211.6	34.3	10.8	10		
NICOTINAMIDE 1X <sup>E</sup>	204.9	15.3	4.8	10	>0.1	-3
NICOTINAMIDE 2X <sup>F</sup>	201.6	13.4	4.2	10	>0.1	-5
NICOTINAMIDE 3X <sup>G</sup>	211.3	17.4	5.5	10	>0.1	0
MICE BEARING MAMMARY TUMORS						
SALINE CONTROL	138.1	33.5	10.6	20		
NICOTINAMIDE 1X	184.5	22.8	5.1	20	<0.0005	+33
NICOTINAMIDE 2X	208.2	27.4	6.1	20	<0.0005	+51
NICOTINAMIDE 3X	204.8	33.2	7.4	20	<0.0005	+48

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE IX. THE EFFECT OF ONE, TWO OR THREE 1 GM/KG DOSES OF NICOTINAMIDE ON THE  
 (CONTINUED) CONCENTRATION OF PFOB (MG/G) IN THE SPLEEN 48 HOURS AFTER THE  
 ADMINISTRATION OF PFOB

NORMAL HEALTHY MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	211.6	34.3	10.8	10		
NICOTINAMIDE 1X <sup>E</sup>	204.9	15.3	4.8	10	>0.1	-3
NICOTINAMIDE 2X <sup>F</sup>	201.6	13.4	4.2	10	>0.1	-5
NICOTINAMIDE 3X <sup>G</sup>	211.3	17.4	5.5	10	>0.1	0
MICE BEARING COLON TUMORS						
SALINE CONTROL	165.1	32.2	10.2	10		
NICOTINAMIDE 1X	146.6	30.4	9.6	10	>0.1	-11
NICOTINAMIDE 2X	187.4	23.0	7.2	10	<0.05	+14
NICOTINAMIDE 3X	195.8	18.1	5.7	10	<0.01	+19

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE).

E 1X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

F 2X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE AND 3.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

G 3X REFLECTS THAT ONE DOSE WAS ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 23.5 HOURS AFTER THE ADMINISTRATION OF PFOB INTRAVENOUSLY.

TABLE X. THE EFFECT OF THREE DOSES OF A VASCULAR DWELL-TIME ENHANCING AGENT ON THE CONCENTRATION OF PFOB (MG/G) IN THE BLOOD 48 HOURS AFTER THE ADMINISTRATION OF PFOB.<sup>1</sup>

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	%CHANGE
10	SALINE CONTROL	18.21	6.2	1.42	19	
	NICOTINAMIDE (500 MG/KG)	25.12	5.88	1.28	21	0.005
	ISONIAZID (10 MG/KG)	19.73	6.57	1.47	20	0.15
15	NEOMYCIN (5 MG/KG)	19.94	7.41	1.62	21	+10
MICE WITH MAMMARY TUMORS						
SALINE CONTROL	3.11	3.84	1.03	14		
NICOTINAMIDE (500 MG/KG)	7.82	5.55	1.48	14	0.008	
ISONIAZID (10 MG/KG)	2.61	1.71	0.44	15	-16	

25

1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE INTRAPERITONEALLY ADMINISTERED 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

30 B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE."

SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE

35 USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, NEOMYCIN AND ISONIAZID).

TABLE XI. THE EFFECT OF THREE DOSES OF A VASCULAR DWELL-TIME ENHANCING AGENT ON THE CONCENTRATION OF PFOB (MG/G) IN THE SPLEEN 48 HOURS AFTER THE ADMINISTRATION OF PFOB.<sup>1</sup>

NORMAL MICE							
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE	
SALINE CONTROL	174.6	23.15	5.01	22			
NICOTINAMIDE (500 MG/KG)	190	20.97	4.47	22	0.014	+9	
ISONIAZID (10 MG/KG)	174.7	32.32	4.47	22	0.15	+0	
NEOMYCIN (5 MG/KG)	170.1	19.6	4.18	22	0.15	-3	
MICE WITH MAMMARY TUMORS							
SALINE CONTROL	154.8	12.4	3.3	14			
NICOTINAMIDE (500 MG/KG)	167.3	20.9	5.58	14	0.033	+8	
ISONIAZID (10 MG/KG)	161.4	14	3.62	15	0.096	+4	

25

1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE INTRAPERITONEALLY ADMINISTERED 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

30 B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE

35 USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, NEOMYCIN AND ISONIAZID).

TABLE XII. THE EFFECT OF THREE DOSES OF A VASCULAR DWELL-TIME ENHANCING AGENT ON THE CONCENTRATION OF PFOB (MG/G) IN THE LIVER 48 HOURS AFTER THE ADMINISTRATION OF PFOB.<sup>1</sup>

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
10	SALINE CONTROL	35.08	7.22	1.54	22	
	NICOTINAMIDE (500 MG/KG)	28.6	5.33	1.14	22	0.001
	ISONIAZID (10 MG/KG)	33.22	5.65	1.2	22	0.15
15	NEOMYCIN (5 MG/KG)	32.66	5.91	1.29	22	0.115
MICE WITH MAMMARY TUMORS						
SALINE CONTROL	39.88	4.42	1.18	14		
NICOTINAMIDE (500 MG/KG)	37.84	3.8	1.02	14	0.1	
20	ISONIAZID (10 MG/KG)	42.79	5.68	1.47	15	0.069

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1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE INTRAPERITONEALLY ADMINISTERED 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

30 B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE."

SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE 35 USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, NEOMYCIN AND ISONIAZID).

TABLE XIII. THE EFFECT OF THREE DOSES OF A VASCULAR DWELL-TIME ENHANCING AGENT ON THE CONCENTRATION OF PFOB (MG/G) IN THE LUNG 48 HOURS AFTER THE ADMINISTRATION OF PFOB.<sup>1</sup>

NORMAL MICE						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	28.45	7.84	1.67	22		
NICOTINAMIDE (500 MG/KG)	22.04	7.08	1.51	22	0.003	-23
ISONIAZID (10 MG/KG)	28.19	8.79	1.87	22	0.15	-1
NEOMYCIN (5 MG/KG)	29.27	8.79	1.87	22	0.15	3
MICE WITH MAMMARY TUMORS						
SALINE CONTROL	24.91	6.56	1.75	14		
NICOTINAMIDE (500 MG/KG)	15.35	2.36	.063	14	0.005	-39
ISONIAZID (10 MG/KG)	26.56	3.15	0.81	15	0.15	7

1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE INTRAPERITONEALLY ADMINISTERED 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

30 B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, NEOMYCIN AND ISONIAZID).

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TABLE XIV. THE EFFECT OF THREE DOSES OF A VASCULAR DWELL-TIME ENHANCING AGENT ON THE CONCENTRATION OF PFOB (MG/G) IN A MAMMARY TUMOR 48 HOURS AFTER THE ADMINISTRATION OF PFOB.<sup>1</sup>

MICE WITH MAMMARY TUMORS						
	MEAN	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
	SALINE CONTROL	6.98	1.75	0.47	14	
10	NICOTINAMIDE (500 MG/KG)	6.01	1.59	0.43	14	0.069
	ISONIAZID (10 MG/KG)	5.87	1.06	.027	15	0.023

15

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- 25 1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE INTRAPERITONEALLY ADMINISTERED 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

30 C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, NEOMYCIN AND ISONIAZID).

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TABLE XV. THE EFFECT OF THREE DOSES OF IBUPROFEN ON THE CONCENTRATION OF PFOB (MG/G) IN BLOOD 48 HOURS AFTER THE ADMINISTRATION OF PFOB.

NORMAL MICE					
	MEAN	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>	% CHANGE
SALINE CONTROL	9.38	0.71	19		
IBUPROFEN	18.56	3.61	18	<.01	49
MICE WITH MAMMARY TUMORS					
SALINE CONTROL	0.34	0.71	19		
IBUPROFEN	1.94	3.61	19	<.05	83
MICE WITH LEWIS LUNG TUMORS					
SALINE CONTROL	1.67	0.71	19		
IBUPROFEN	4.49	3.61	18	<.05	63

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25

1 IBUPROFEN WAS ADMINISTERED INTRAPERITONEALLY 24 HOURS BEFORE, 30 MINUTES BEFORE AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./N

30 C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS IBUPROFEN).

Results and Discussion

The results of Experiment 1 demonstrate that, as the dosage of nicotinamide increased, the 5 concentration of PFOB within the blood increased as well. With increasing dosages of nicotinamide, decreasing amounts of PFOB concentrated within the bone marrow, liver and lung. Finally, neomycin, in comparison to nicotinamide, was able to increase the concentration of 10 PFOB in the blood of tumor bearing mice to a greater extent. The difference in efficacies of neomycin and nicotinamide, however, was not statistically significant.

The results of Experiment 2 demonstrate that, with successive doses of nicotinamide, the concentration 15 of PFOB within the blood increased. These effects were most pronounced in mice with mammary or colon tumors.

The concentration of PFOB in the blood of tumor bearing mice was lower than the concentration of PFOB in the blood of normal mice. This depressed level of PFOB 20 in the blood of tumor bearing mice is believed to be due to the fact that the tumors themselves had activated the macrophages of the liver, bone marrow and lung. Accordingly, this priming of the macrophages had enhanced phagocytosis as well as the production of ecoisanoids and 25 cytokines -- even before the administration of PFOB.

With respect to normal mice, an increase in the concentration of PFOB in the blood was coupled with a decrease in the concentration of PFOB within the spleen, liver, lung and bone marrow. In mice bearing tumors, 30 however, the concentration of PFOB in the spleen also increased as the concentration of PFOB increased in the blood.

The observed lower levels of PFOB in the spleen of tumor-bearing mice are thought to be due to the 35 suppression of spleen phagocytes. This phenomenon is thought to be due to the enhanced production of cytokines and ecoisanoids by activated macrophages located

elsewhere in the reticuloendothelial system of tumor-bearing mice. Accordingly, it is believed that phagocytosis in the spleen increased in tumor bearing mice following the administration of nicotinamide because 5 nicotinamide down-regulated the production of cytokines and eicosanoids by these other macrophages.

The spleen is an important organ in the immunosuppression against infection. Inhibition of the splenic phagocytes in tumor-bearing mice may contribute 10 to these subjects' lowered resistance to infection. Thus, the administration of megadoses of vascular dwell-time enhancing agents, like nicotinamide, may be useful in overcoming the general immunosuppression seen in subjects with malignant tumors.

15 The results of Experiment 3 demonstrate that different vascular dwell-time enhancing agents, at different doses, were capable of increasing the concentration of PFOB within the blood while simultaneously lowering the PFOB concentration within 20 other organs, such as the liver. These effects were more pronounced in mice bearing mammary tumors as compared to normal mice. Of the four agents tested, nicotinamide, at its dose, was the most effective agent.

The results of Experiment 4 demonstrate that 25 ibuprofen is also capable of increasing the concentration of PFOB within the blood. As noted previously, the depressed levels of PFOB in the blood of tumor bearing mice is believed to be due to the priming of macrophage activity by the tumors present in these mice. 30 Furthermore, differences in the levels of PFOB in the blood of mice bearing different types of tumors is believed to reflect that different tumors induce different levels of macrophage activity and, hence, different levels of phagocytosis.

### Conclusion

It has been demonstrated that vascular dwell-time enhancing agents, such as nicotinamide, isoniazid, neomycin and ibuprofen are capable of extending the dwell-time of particulate therapeutic and diagnostic agents, such as PFOB, within the vascular compartment. These results suggest that nicotinamide, isoniazid, neomycin and ibuprofen, as well as other vascular dwell-time enhancing agents, are capable of enhancing the efficacy of a particulate therapeutic or a particulate diagnostic agent by allowing the latter to remain within the vascular compartment or to be carried within the vascular compartment to a target.

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### EXAMPLE II

#### The Effects of Nicotinamide, Isoniazid and Neomycin on Weight in Mammals Receiving PFOB

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One experiment was conducted in order to assess the ability of different vascular-dwell time enhancing agents to prevent weight loss normally induced by the administration of a particulate therapeutic or a particulate diagnostic agent. In particular, this experiment focused on the effects of three agents, nicotinamide, isoniazid and neomycin, on the weight of mice being administered PFOB.

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### Materials and Methods

#### Mammals:

Female Balb/C normal healthy mice as well as female Balb/C mice bearing mammary tumors were employed in this study. Each mouse weighed approximately 20 gm.

Drugs:

Perfluorooctylbromide (PFOB) was procured from Alliance Pharmaceutical Corp. and administered intravenously as an emulsion (90 gm/100 ml) in the tail vein.

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Nicotinamide (Sigma), isoniazid (Sigma), and neomycin (Sigma) solutions were freshly prepared before each experiment. These compounds were dissolved in water and administered intravenously.

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Normal Saline (0.9% w/v) was administered intravenously in the tail vein.

Weight Measurements:

The weight of each mouse was recorded prior to 15 the administration of PFOB as well as 48 hours after the administration of PFOB.

## EXPERIMENT 1

Mice were administered three bolus doses 20 intraperitoneally of a vascular dwell-time enhancing agent. Nicotinamide was administered at a dose of 500 mg/kg, isoniazid was administered at a dose of 10 mg/kg and neomycin was administered at a dose of 5 mg/kg. Forty-eight hours post injection, the mice were 25 sacrificed by cervical dislocation. The results of this experiment are summarized in Tables XVI to XVII.

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TABLE XVI. THE EFFECT OF NICOTINAMIDE, ISONIAZID AND NEOMYCIN ON THE BODY WEIGHT (G) OF NORMAL MICE 48 HOURS AFTER THE ADMINISTRATION OF PFOB

	MEAN AT DAY 0	NET CHANGE AT DAY 2	% CHANGE AT DAY 2	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>
5	SALINE CONTROL	19.89	-0.41	-1.85	0.83	0.19	19
10	NICOTINAMIDE (500 MG/KG)	19.4	.00	0.00	0.62	0.14	21 <0.05
	ISONIAZID (10 MG/KG)	19.66	-0.71	-3.21	0.85	0.19	>0.1
	NEOMYCIN (5 MG/KG)	19.81	0.48	2.17	0.30	0.17	>0.1

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25 1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE ADMINISTERED INTRAPERITONEALLY 30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

30 C N REFERS TO THE NUMBER OF MICE TESTED.

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE ALONE." SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS. 1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT

35 T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER CATEGORIES OF THERAPY (SUCH AS NICOTINAMIDE, ISONIAZID AND NEOMYCIN).

TABLE XVII. THE EFFECT OF NICOTINAMIDE AND ISONIAZID ON THE BODY WEIGHT (G) OF MICE BEARING MAMMARY TUMORS 48 HOURS AFTER THE ADMINISTRATION OF PFOB

	MEAN AT DAY 0	NET CHANGE AT DAY 2	% CHANGE AT DAY 2	S.D. <sup>A</sup>	S.E. <sup>B</sup>	N <sup>C</sup>	P VALUE <sup>D</sup>
5	SALINE CONTROL	22.11	-0.03	-0.14	0.41	0.11	14
	NICOTINAMIDE (500 MG/KG)	22.99	0.46	2.08	0.69	0.18	14 <0.025
	ISONIAZID (10 MG/KG)	21.87	-0.17	-0.77	0.42	0.11	15 >.1

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25 1 THE VASCULAR DWELL-TIME ENHANCING AGENTS WERE ADMINISTERED INTRAPERITONEALLY

30 MINUTES BEFORE, 3.5 HOURS AFTER AND 24 HOURS AFTER THE INTRAVENOUS  
ADMINISTRATION OF PFOB.

A S.D. REFERS TO THE STANDARD DEVIATION OF THE MEAN

B S.E. REFERS TO THE STANDARD ERROR OF THE MEAN. S.E. = S.D./ $\sqrt{N}$

30 C N REFERS TO THE NUMBER OF MICE TESTED

D P VALUE REFERS TO "THE PROBABILITY OF OBTAINING A RESULT AS EXTREME AS OR MORE  
EXTREME THAN THE ONE OBSERVED IF THE DISSIMILARITY IS ENTIRELY DUE TO CHANCE  
ALONE. SEE, MEDICAL USES OF STATISTICS (J.C. BAILOR, III AND F. MOSTETLER, EDS.  
1986). THE USUAL SIGNIFICANCE LEVEL OF 0.05 IS USED WITH A ONE-SIDED STUDENT

35 T-TEST COMPARING A SALINE CONTROL THERAPY WITH THE RESULTS OF THE OTHER  
CATEGORIES OF THERAPY UNDER INVESTIGATION (SUCH AS NICOTINAMIDE, ISONIAZID AND  
NEOMYCIN).

What is claimed is:

1. A method for delivering a particulate therapeutic agent or a particulate diagnostic agent by means of the vascular compartment of a mammal which comprises the adjunct administration of an effective amount of said particulate therapeutic agent or particulate diagnostic agent and an effective amount of a vascular dwell-time enhancing agent to said mammal, with the proviso that said particulate therapeutic agent or particulate diagnostic agent is not a particulate blood substitute.
2. The method of claim 1 wherein said particulate therapeutic agent or particulate diagnostic agent measures from about .001 microns to about 50 microns in diameter.
3. The method of claim 1 wherein said particulate therapeutic agent is selected from the group consisting of a particulate chemotherapeutic agent, a particulate vasopressor, a particulate anti-inflammatory agent, a particulate anesthetic, a particulate vasodilator, a particulate tranquilizer, a particulate soporific and a particulate sedative or said particulate diagnostic agent is selected from the group consisting of a particulate antibody and a particulate radiolabelled compound.
4. The method of claim 1 wherein said particulate therapeutic agent or particulate diagnostic agent is associated with a carrier system.
5. The method of claim 4 wherein said carrier system is selected from the group consisting of a lipid,

a liposome, a microsphere, a molecule, an artificial cell, a microcapsule or a microcarrier bead.

6. The method of claim 1 wherein said  
5 vascular dwell-time enhancing agent is associated with a carrier system.

7. The method of claim 6 wherein said carrier system is a molecule.

10 8. A method of delivering a particulate blood substitute by means of the vascular compartment of a mammal which comprises the adjunct administration of an effective amount of said particulate blood substitute and  
15 an effective amount of a vascular dwell-time enhancing agent to said mammal.

9. A method for preventing or treating a mammalian condition selected from the group consisting of  
20 hypoxia in non-neoplastic cells, hypoxemia and anemia which comprises the adjunct administration of a therapeutically effective amount of a particulate blood substitute and an effective amount of a vascular dwell-time enhancing agent to said mammal in need of said  
25 prevention or treatment.

10. A method treating a neoplastic condition in a mammal which comprises irradiating a tumor cell in said mammal at least about three hours after the adjunct  
30 administration of a therapeutically effective amount of a particulate blood substitute and an effective amount of a vascular dwell-time enhancing agent to said mammal.

11. The method of claim 10 wherein said tumor  
35 cell is irradiated at about 48 hours or at about 72 hours after the adjunct administration of a therapeutically

effective amount of said particulate blood substitute and an effective amount of said vascular dwell-time enhancing agent.

5           12. The method of claim 8, 9 or 10 wherein said particulate blood substitute is associated with a carrier system.

10          13. The method of claim 12 wherein said carrier system is selected from the group consisting of a lipid, a liposome, a microsphere, a molecule, an artificial cell, a microcapsule or a microcarrier bead.

14. The method of claim 8, 9 or 10 wherein  
15 said particulate blood substitute is selected from the group consisting of a hemoglobin based composition and a fluorocarbon.

16. The method of claim 8, 9 or 10 wherein  
20 said particulate blood substitute is selected from the group consisting of polymerized hemoglobin and hemoglobin associated with a carrier system.

17. The method of claim 8, 9 or 10 wherein  
25 said particulate blood substitute measures from about .001 microns to about 50 microns in diameter.

18. A method for imaging a selected tissue or organ of a mammal which comprises imaging said tissue or  
30 organ of said mammal after the adjunct administration of an effective amount of a particulate imaging agent and an effective amount of a vascular dwell-time enhancing agent to said mammal.

18. The method of claim 17 wherein said method of imaging is selected from the group consisting of ultrasound imaging, X-ray imaging and MRI.

5           19. The method of claim 17 wherein said tissue or organ is selected from the group consisting of the heart, blood, a part of the lymphatic system, a part of the reticuloendothelial system, the liver, lung, spleen and bone marrow.

10           20. The method of claim 17 wherein said particulate contrast agent measures from about .001 microns to about 50 microns in diameter.

15           21. The method of claim 17 wherein said particulate imaging agent is associated with a carrier system.

20           22. The method of claim 21 wherein said carrier system is selected from the group consisting of a lipid, a liposome, a microsphere, a molecule, an artificial cell, a microcapsule or a microcarrier bead.

25           23. The method of claim 17 wherein said method of imaging is ultrasound imaging and said particulate contrast agent is selected from the group consisting of a proteinaceous matrix containing oxygen and a fluorocarbon.

30           24. The method of claim 17 wherein said particulate contrast agent is an albumin matrix containing oxygen.

35           25. The method of claim 17 wherein said particulate contrast agent is selected from the group consisting of a compound labeled with a heavy halide.

26. The method of claim 17 wherein said method of imaging is X-ray imaging and said particulate contrast agent is selected from the group consisting of iodine, bromine, 1-bromoheptadecafluoroctane,  
5 1-bromopentadecafluoroheptane, 1-bromotridecafluorohexane,  $C_6F_{12}Br_2$ ,  $C_7F_{14}Br_2$  and  $C_8F_{16}Br_2$ .

27. The method of claim 17 wherein said particulate contrast agent is selected from the group  
10 consisting of a monobrominated perfluorocarbon, a dibrominated perfluorocarbon, a monoiodinated perfluorocarbon and a di-iodinated perfluorocarbon.

28. The method of claim 17 wherein said method  
15 of imaging is MRI and said particulate contrast agent is selected from the group consisting of a paramagnetic compound, a ferromagnetic compound and a superparamagnetic compound.

20 29. The method of claim 17 wherein said particulate contrast agent is selected from the group consisting of ferric oxyhydroxide, gadolinium oxide, magnetite, ferrite, gamma ferric oxide, superparamagnetic iron oxide and a fluorocarbon.

25 30. The method of claim 14, 22 or 28 wherein said fluorocarbon is selected from the group consisting of a bis(F-alkyl)ethane, a cyclic fluorocarbon, a perfluorinated amine, a halogenated perfluorocarbon, a  
30 perfluoroalkylated ether, a perfluoroalkylated polyether, a perfluorocyclic ether, a perfluoroalkylated hydride and a fluorocarbon-hydrocarbon.

35 31. The method of claim 14, 22 or 28 wherein said fluorocarbon is selected from the group consisting of  $C_4F_9CH=CHC_4F_9$ ,  $i-C_3F_7CH=CHC_6F_{13}$ ,  $C_6F_{13}CH=CHC_6F_{13}$ ,

$C_3F_7CBr=CBuC_3F_7$ ,  $C_3F_7Cl=CIC_3F_7$ ,  $C_2F_5CBr=CBuC_2F_5$ ,  $C_2F_5Cl=CIC_2F_5$ ,  
 $C_{10}F_{18}$ , F-adamantane, F-methyladamantane,  
F-1,3-dimethyladamantane, F-dimethylbicyclo[3,3,1]nonane,  
F-tri-methylbicyclo[3,3,1]nonane, F-tripropylamine, F-  
5 tributylamine, F-4-methyloctylhydroquinolizine,  
F-n-methyl-decahydroisoquinoline,  
F-n-cyclohexylpyrrolidine, F-2-butyltetrahydrofuran,  
1-bromoheptadecafluoroctane,  
1-bromopentadecafluoroheptane,  
10 1-bromotridecafluorohexane,  $C_6F_{12}Br_2$ ,  $C_7F_{14}Br_2$ ,  $C_8F_{16}Br_2$ ,  
 $(CF_3)_2CFO(CF_2CF_2)_3F$ ,  $(C_6F_{13})_2O$ ,  $F[CF(CF)CFO]CHFCF$ ,  
 $(CF_3)_2CFO(CF_2CF_2)_2OCF(CF_3)_2$ ,  $(CF_3)_2CFO(CF_2CF_2)_3OCF(CF_3)$ ,  
perfluoroctylhydride, perfluorononylhydride,  
perfluorodecylhydride,  $C_8F_{17}C_2H_5$ ,  $C_6F_{13}CH=CHC_6H_{13}$ , and  $C_8F_{17}R$ ,  
15 wherein R is any hydrocarbon with an organic functional  
group.

32. The method of claim 14, 22 or 28 wherein  
said fluorocarbon is selected from the group consisting  
20 of a monobrominated perfluorocarbon, a monoiodinated  
perfluorocarbon, a dibrominated perfluorocarbon and a di-  
iodinated pefluorocarbon.

33. The method of claim 1, 5, 9, 10 or 17  
25 wherein said vascular dwell-time enhancing agent is  
selected from the group consisting of a lipid metabolism  
modifying agent, an antibiotic, a cyclooxygenase  
inhibitor, a NADase inhibitor, an ADP-ribose-polymerase  
inhibitor and an ADP-ribose-synthetase inhibitor.

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34. The method of claim 1, 5, 9, 10 or 17  
wherein said vascular dwell-time enhancing agent is  
selected from the group consisting of clofibrate,  
gemfibrizol, probucol, insulin, thyroxin, lovastatin,  
35 nicotinamide, nicotinic acid, an antibiotic derived from  
Streptomyces, an antibiotic derived from Bacillus

- subtilis, an antibiotic derived from Cephalosporin acremonium, an antibiotic derived from Microspora purpurea, an antibiotic derived from Micromonospora echinospora an antibiotic derived from a variant of  
5 Micromonospora purpurea and an antibiotic derived from a variant of Micromonospora echinospora.

35. The method of claim 1, 5, 9, 10 or 17 wherein said vascular dwell-time enhancing agent is  
10 selected from the group consisting of neomycin, streptomycin, kanamycin, paromomycin, chloramphenicol, clindamycin, cefoxitin, amphotericin B, tobramycin, amikacin, bacitracin, cephalosporin C, a semisynthetic antibiotic derivative of cephalosporin C, gentamicin,  
15 ibuprofen, benzamide, a benzamide analog, a nicotinamide analog, theophylline, thymine and a thymine analog.

36. The method of claim 1, 5, 9, 10 or 17 wherein said vascular dwell-time enhancing agent is  
20 selected from the group consisting of 3-aminobenzamide, benzoic acid, 3-aminobenzoic acid,  $\alpha$ -amino-3-indolepropionic acid, isoniazid, NAD, NADH, NADPH, thymine riboside and thymidine.

25 37. The method of claim 1, 5, 9, 10 or 17 wherein said vascular dwell-time enhancing agent is associated with a carrier system.

30 38. The method of claim 37 wherein said carrier system is a molecule.

39. The method of claim 1, 5, 9, 10 or 17 wherein said mammal is a human.

35 40. A method for preventing or treating cachexia in a mammal which comprises the administration

of a therapeutically effective amount of an anti-cachexia agent to a mammal in need of said prevention or treatment.

5           41. The method of claim 40 wherein said anti-cachexia agent is selected from the group consisting of an ADP-ribose-polymerase inhibitor, an ADP-ribose transferase inhibitor and a NADase inhibitor.

10           42. The method of claim 40 wherein said anti-cachexia agent is selected from the group consisting of nicotinamide, a nicotinamide analog, benzamide, a benzamide analog, theophylline, thymine, a thymine analog.

15           43. The method of claim 40 wherein said anti-cachexia agent is selected from the group consisting of nicotinic acid, 3-aminobenzamide, benzoic acid, 3-aminobenzoic acid,  $\alpha$ -amino-3-indolepropionic acid, NAD, 20 NADH, NADPH, thymine riboside and thymidine.

44. The method of claim 40 wherein said anti-cachexia agent is associated with a carrier system.

25           45. The method of claim 44 wherein said carrier system is a molecule.

46. A composition comprising a vascular dwell-time enhancing agent and a particulate therapeutic or a 30 particulate diagnostic agent.

47. The composition of claim 44 wherein said vascular dwell-time enhancing agent is selected from the group consisting of a lipid metabolism modifying agent, 35 an antibiotic, a cyclooxygenase inhibitor, a NADase

inhibitor, an ADP-ribose-polymerase inhibitor, and an ADP-ribose-synthetase inhibitor.

48. The composition of claim 37 wherein said  
5 vascular dwell-time enhancing agent is selected from the group consisting of clofibrate, gemfibrizol, probucol, insulin, thyroxin, lovastatin, nicotinamide, nicotinic acid, an antibiotic derived from Streptomyces, an antibiotic derived from Bacillus subtilis, an antibiotic  
10 derived from Cephalosporin acremonium, an antibiotic derived from Microspora purpurea, an antibiotic derived from Micromonospora echinospora, an antibiotic derived from a variant of Micromonospora purpurea and an antibiotic derived from a variant of Micromonospora  
15 echinospora

49. The composition of claim 40 wherein said vascular dwell-time enhancing agent is selected from the group consisting of neomycin, streptomycin, kanamycin,  
20 paromomycin, chloramphenicol, clindamycin, cefoxitin, amphotericin B, tobramycin, amikacin, bacitracin, cephalosporin C, a semisynthetic antibiotic derivative of cephalosporin C, gentamicin, ibuprofen, a benzamide, a benzamide analog, a nicotinamide analog, theophylline,  
25 thymine and a thymine analog.

50. The composition of claim 40 wherein said vascular dwell-time enhancing agent is selected from the group consisting of 3-aminobenzamide, benzoic acid,  
30 3-aminobenzoic acid,  $\alpha$ -amino-3-indolepropionic acid, isoniazid, NAD, NADH, NADPH, thymine riboside and thymidine.

51. The composition of claim 40 wherein said  
35 vascular dwell-time enhancing agent is associated with a carrier system.

52. The composition of claim 51 wherein said carrier system is a molecule.

53. The composition of claim 40 wherein said particulate therapeutic agent is selected from the group consisting of a particulate hemoglobin based composition, a particulate chemotherapeutic agent, a particulate vasopressor, a particulate anti-inflammatory agent, a particulate anesthetic, a particulate vasodilator, a particulate tranquilizer, a particulate soporific, a particulate sedative, a polymerized hemoglobin and hemoglobin associated with a carrier system or said particulate diagnostic agent is selected from the group consisting of a particulate antibody and a particulate radiolabelled compound.

54. The composition of claim 40 wherein said particulate therapeutic agent or particulate diagnostic agent is a fluorocarbon.

55. The composition of claim 40 wherein said fluorocarbon is selected from the group consisting of a bis(F-alkyl)ethane, a cyclic fluorocarbon, a perfluorinated amine, a halogenated perfluorocarbon, a perfluoroalkylated ether, a perfluoroalkylated polyether, a perfluorocyclic ether, a perfluoroalkylated hydride and a fluorocarbon-hydrocarbon.

56. The composition of claim 40 wherein said fluorocarbon is selected from the group consisting of  $C_4F_9CH=CHC_4F_9$ ,  $i-C_3F_7CH=CHC_6F_{13}$ ,  $C_6F_{13}CH=CHC_6F_{13}$ ,  $C_3F_7CBr=CBrC_3F_7$ ,  $C_3F_7Cl=CIC_3F_7$ ,  $C_2F_5CBr=CBrC_2F_5$ ,  $C_2F_5Cl=CIC_2F_5$ ,  $C_{10}F_{18}$ , F-adamantane, F-methyladamantane, F-1,3-dimethyladamantane, F-dimethylbicyclo[3,3,1]nonane, F-tri-methylbicyclo[3,3,1]nonane, F-tripropylamine, F-tributylamine, F-4-methyloctylhydroquinolizine,

- F-n-methyl-decahydroisoquinoline,  
F-n-cyclohexylpyrrolidine, F-2-butyltetrahydrofuran,  
1-bromoheptadecafluoroctane,  
1-bromopentadecafluoroheptane,  
**5** 1-bromotridecafluorohexane,  $C_6F_{12}Br_2$ ,  $C_7F_{14}Br_2$ ,  $C_8F_{16}Br_2$ ,  
 $(CF_3)_2CFO(CF_2CF_2)_3F$ ,  $(C_6F_{13})_2O$ ,  $F[CF(CF)CFO]CHFCF$ ,  
 $(CF_3)_2CFO(CF_2CF_2)_2OCF(CF_3)_2$ ,  $(CF_3)_2CFO(CF_2CF_2)_3OCF(CF_3)$ ,  
perfluoroctylhydride, perfluorononylhydride,  
perfluorodecylhydride,  $C_8F_{17}C_2H_5$ ,  $C_6F_{13}CH=CHC_6H_{13}$  and  $C_8F_{17}R$   
**10** wherein R is any hydrocarbon with an organic functional group.

**57.** The composition of claim 40 wherein said fluorocarbon is selected from the group consisting of a  
**15** monobrominated perfluorocarbon, a moniodinated perfluorocarbon, a dibrominated perfluorocarbon and a di-iodinated perfluorocarbon,

**58.** The composition of claim 40 wherein said  
**20** particulate therapeutic agent or particulate diagnostic agent is associated with a carrier system.

**59.** The method of claim 58 wherein said carrier system is selected from the group consisting of a lipid, a liposome, a microsphere, a molecule, an  
**25** artificial cell, a microcapsule or a microcarrier bead.

**60.** A kit comprising:  
**30** i) a vascular dwell-time enhancing agent associated with a carrier system; and  
ii) either a particulate therapeutic agent or a particulate diagnostic agent.